Indian Journal of Science and Technology, *Vol 8(32), DOI: 10.17485/ijst/2015/v8i32/87755, November 2015*
ISSN (Print) : 0974-6846
ISSN (Online) : 0974-5645

Study on Methanol Production with Vegetable in Rotating Reactor

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Abstract

Fuel methanol is on demand nowadays. Lignocelluloses and algal biomasses are largely generated as waste materials out of agricultural practises and process industries. In the present work, methanol production by *Saccharomyces cerevisiae* was carried out using lignocellulosic biomass (vegetable) as a substrate and using rotating biological reactor that utilizes biologically pretreated vegetable as substrates for methanol production and its process optimization. During the batch experiment with varying substrate concentration (60-100 g/l), pH (4.8-5.8) and commercial cellulase enzyme concentration of (10- 20 mg/ml) resulted in 38.9 g/l of methanol concentration with a maximum of 17 % yield. The SEM analysis was carried out for analysing the structural morphology of the untreated, pretreated, hydrolysed and fermented samples.

Keywords: Agricultural Waste, Biological Reactor, Lignocellulosic Biomass, Methanol, Optimization, Vegetable

1. Introduction

Lignocellulose consists of lignin, hemicellulose and cellulose which make them a substrate of enormous biotechnological value[4]. It is misfortune that because of complex chemical nature of the lignocelluloses, and algal biomass these abundant resources are considered as recalcitrant molecules which cannot be degraded easily is any means[3]. Response Surface Methodology (RSM) has become an effective method in helping to solve many practical problems in the course of production, such as reducing manufacturing costs, optimizing process operating conditions, and improving product quality. It has been used successfully to model and optimize biochemical processes in agriculture, biology, and chemistry[1]. The objective of the present work was to optimize the process parameters for the production of methanol with low cost vegetable as substrate using Response Surface Methodology.

2. Materials and Methods

2.1 Reactor Design

Existing vertical bioreactor used for solid state fermentation with agitator creates shear stress to the substrate and depletes the rate of mass transfer and heat transfer. The mixing index for substrate and inoculums is very less[1]. This in turn reduces the yield and conversion of substrate to methanol. The surface area for the biochemical reaction to take place is less in the existing vertical reactors used in industries. Hence a rotating biological reactor was designed and fabricated for the production of methanol. This reactor was used for the production with optimized process parameters. The overall design of the reactor aims at enhancing the diffusion and overcome the mass transfer limitations. According to Ficks law

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2.2 Bioconversion of Pretreated Lignocelluloses to Methanol

After the process of delignification of lignocelluloses by biological pretreatment, the substrate can be effectively used for its bioconversion to methanol. This involves the breaking down of cellulose to glucose and further converting it to methanol by the action of microorganisms[2].

2.3 Process Optimization

Process optimization was carried out by Response Surface Methodology using Design-Expert software Version 8.0.3.1. RSM is the product of the combination of mathematics and statistics, which researches and optimizes the relationship between factors and response value A 2^3 full factorial experimental design with 14 experiments were employed which includes 8 trails for each axial point and 6 trails for replication of the central points based on the pattern generated through software[11–13]. Experiment was conducted as per the design matrix in 250ml Erlenmeyer flasks with varying amount of substrate, inoculums and fermentation time. pH (4.8-5.8), temperature (30-40 °C), substrate concentration (60-100g/l of vegetable were the parameters and Methanol yield was taken as the response[3].

2.4 Estimation of Methanol Content

Estimation of methanol content was carried out in the fermented samples pretreated with *Pleurotus florida* . The method used for the estimation of methanol content is Chromic Acid Assay.

The methanol yield is analyzed using formula:

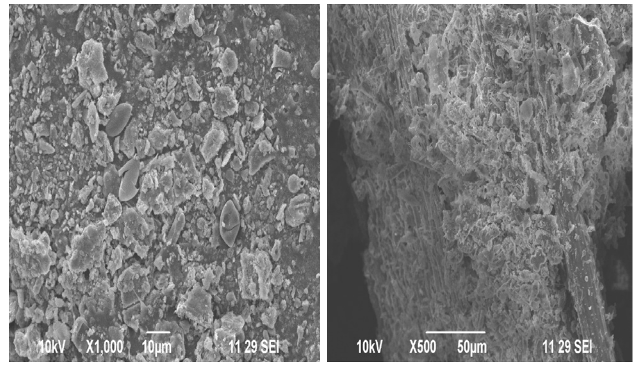
Yield % = E *0.5/S *0.9

Where E - Methanol concentration (g/l)

S - Cellulose concentration (g/l).

3. Results and Discussions

The Biologically pre-treated samples were analysed for their structural characteristics by Scanning Electron Microscopy.The size of the vegetable treated with *Pleurotus florida* was approximately 100μm and it was reduced to many ground fibres (Figure 1). The vegetable treated with *Pleurotus florida* and grape leaves was gradually reduced to fine ground particles, thus creating particles with large surface area[4,5].



(a) (b)

Figure 1. **(a)** Rice straw untreated. **(b)** Vegetable pre-treated.

3.1 Optimisation of Process Conditions for Vegetable and Algal Biomass

3.1.1 Response Surface Methodology

Central composite design was employed to study the interactions between the most significant variables, namely amount of substrate, enzyme concentration and Ph. The design matrix of tested variables and the experimental results are represented in Table 1. It was observed that Run 7 gives the highest percentage of methanol (17.97%) and concentration as 38.9g/l as the response[6].

The adequacy of the model was checked using ANOVA, as shown in the Table 2. The Model F-value of

Table 1. Experimental designs and results of central composite design

Source	Sum of squares	Df	Mean square	F-value	P-value, Prob>F
Model		9	31.39	43	0.0001
A	0.091	1	63.93	89	0.0001
B	0.023	1	5.02	7	0.0001
C	0.19	1	86.76	121	0.0382
AB	1.575E-004	1	11.34	15	0.001
AC	0.085	1	8.91	12	0.0124
BC	0.071	1	39.63	55	0.0003
A^2	0.12	1	4.12	5	0.0536
B^2	0.028	1	4.3	10.08	0.0099
C^2	0.032	1	0.00		0.0067
Residual	0.028	10	0.0000		
Lack of fit	0.021	5			0.1317
Pure error	7.076E-003	5			
Total	0.65	19			

43 implies the model is significant. There is only a 0.01% chance that a "Model F-Value" this large could occur due to noise. Values of "Prob > F" less than 0.0500 indicate model terms are significant. In this case A, B, C, AC, BC, A^2, B^2, C^2 are significant model terms. Values greater than 0.1000 indicate the model terms are not significant. The coefficient of determination (R^2) was calculated as 0.9808 for methanol production (section 3.1.2), indicating good agreement between the experimental and the predicted values (Figure 2).

3.1.2 Regression Co-efficient

Std. Dev	**0.85**	**R^2**	**0.9808**
Mean	**8.70**	**Adj R^2**	**0.9584**
C.V.	**9.13**	**Pred R^2**	**0.2285**
PRESS	**273.15**	**Adeq Precision**	**22.362**

A - Amount of substrate, B - Inoculums concentration, C - Fermentation time.

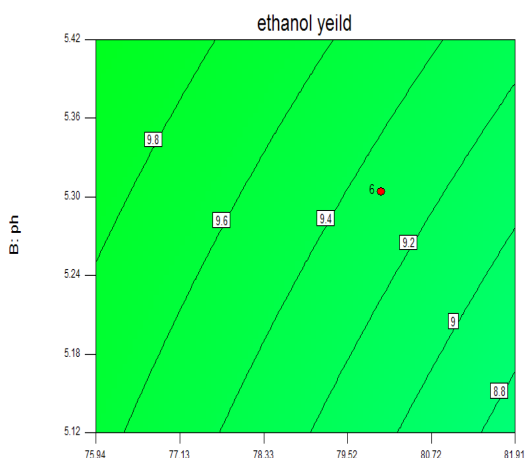


Figure 2. Interaction between substrate concentration and pH on methanol yield using contour and 3D Surface plots.

Thus substrate concentration of 60g/l. Ph of 5.3 and enzyme concentration of 20 mg/g substrate resulted in higher yield of 17.3% and methanol concentration of 38.9 g/l.

The present work focuses on effective pre-treatment and optimization of process parameters for methanol production using low cost substrate (vegetable). Further studies aims at methanol production using the optimized process parameters in the designed reactor[7–10].

4. Conclusion

By effective pre-treatment technology and optimization technique, lignocelluloses biomasses can be utilized as a potential substrate for methanol production and also contribute towards the low cost of production.

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